REMARKS AND ARGUMENTS

Claims 1-30 are presently pending in the application. Claims 1, 5, 6, 13, and 21 are amended without any prejudice or disclaimer of any previously claimed subject matter. Applicant reserves the right to prosecute any cancelled subject matter in a divisional or continuation application. New claims 28-30 are directed to specific embodiments of the present invention. Support for such embodiments can be found on pages 12-14 of the specification.

Rejections under 35 U.S.C. § 112, First Paragraph

The specification and original claims 1-3, 7-15, 19-23, and 27 were rejected under 35 U.S.C. §112, first paragraph, allegedly because the criteria for determining which compounds are effective inosine monophosphate dehydrogenase (IMPDH) inhibitors involves undue experimentation. Applicants respectfully disagree. The Office Action appears to present the position, now disfavored by the U.S. Patent and Trademark Office, that claims should be restricted to the embodiment of presented working examples. This is not the law. In fact, there is no requirement that a patent application contain any working examples whatsoever.

The "undue experimentation" or "Foreman" factors from In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988), while normally used for enablement rejections under 35 U.S.C. 112, first paragraph, are particularly apropos in this instance. In Wands, the Court reversed the U.S.P.T.O.'s rejection that claims for detecting Hepatitis B surface antigens were non-enabling (Id., 8 USPQ2d at 1407). "The Court held that the specification was enabling with respect to the claims at issue and found that 'there was considerable direction and guidance' in the specification; there was a 'high level of skill in the art at the time the application was filed;' and 'all of the methods needed to practice the invention were well known." (MPEP, Sec. 2164.01(a) citing In re Wands , 858 F.2d at 740, 8 USPQ2d at 1407). As in Wands, methods

needed to practice the invention, i.e., to ascertain which compounds are IMPDH inhibitors, and to use such compounds, are well known in the art.

While the Examiner acknowledged that the specification teaches one how to use the particular IMPDH inhibitors set forth in the examples of the present invention, the Examiner alleged that the specification does not teach a skilled artisan how to determine which other compounds are IMPDH inhibitors, and thus useful in the present invention. Applicants respond that the art is replete with references that teach compounds that are effective IMPDH inhibitors as well as various assays to assess the IMPDH activity of a pharmaceutical agent. Numerous IMPDH inhibitors as well as various assays for assessing such activity, are well known in the art, are publicly available, and can be readily accessed. For example, the Examiner is directed to references containing such information – U.S. Patent Nos. 5,380,879; 5,807,876; and 5,932,600 (copies enclosed and cited on the Supplemental Information Disclosure Statement, Form-1449). All of these patents cite numerous IMPDH inhibitors as well as methods to assess IMPDH inhibition based on methodology established in the 1950's and 1960's. Therefore, IMPDH inhibitors and assays to assess which compounds are effective IMPDH inhibitors are well within the purview of the prior art, and well known to a skilled artisan.

Based on the guidance of the specification and the breath of the claim, those of ordinary skill in the art can without undue experimentation ascertain which compounds are effective IMPDH inhibitors in view of knowledge available in the art, including, but not limited to, the U.S. patents cited above.

Applicants submit that, "if a statement of utility in the specification contains within it a connotation of how to use, and/or the art recognizes that standard modes of administration are known and contemplated, 35 U.S.C. 112 is satisfied." (MPEP Sec. 2164.01(c) citing In re Johnson, 282 F.2d 370, 373, 127 U.S.P.Q. 216, 219 (CCPA 1960); In re Hitchings, 342 F.2d 80,87. 144 USPQ 637, 643 (CCPA 1965)). In the instant application, pages 12-14 teach how to use the IMPDH inhibitors of the invention. Suitable modes of administration are disclosed in

detail on pages 23-26 of the specification. In particular, pharmaceutically acceptable compositions for oral, intravenous, parenteral, intradermal, subcutaneous and topical administration are disclosed. Pharmaceutically acceptable dosages have also been disclosed (page 23, lines 20-26), and typically range from about 0.1 mg/kg to about 100 mg/kg per day.

Rejections under 35 U.S.C. § 112, Second Paragraph

Original claims 1 and 4-12 were rejected under 35 U.S.C. §112, second paragraph, allegedly because the term "etherlipidin" is unclear. Applicants have amended the term to "ether lipid in" to correct the minor typographical error.

Rejections under 35 U.S.C. § 102

Original claims 12-27 were rejected under 35 U.S.C. §102, allegedly because U.S. Patent No. 5,270,315 (the '315 patent) to BELLEAU et al., inherently discloses the use of β-D-1,3-dioxolanyl purines for the prophylaxis of HIV. The Examiner alleges that since any medicament used in alternation is considered to include administration of the compounds alone (although sequentially), prophylactic treatments using a known compounds in alternation are inherent.

Without any comment to the Examiner's position, and solely to promote prosecution, Applicants have amended the claims such that the β-D-1,3-dioxolanyl purine and the IMPDH inhibitor are administered in combination, rather than alone. Further, according to the Examiner's suggestion, Applicants have amended the claims such that the host is limited to a host in need thereof.

Rejections under 35 U.S.C. § 103

Original claims 1-27 were rejected under 35 U.S.C. §103, allegedly because they are unpatentable over ICHIMURA et al. "Polymerase Substrate Depletion: A Novel Strategy for Inhibiting the Replication of the Human Immunodeficiency Virus" Virology, **1995**, *211* (2), 554-560; FERNANDEZ-LARSSON et al. "Ribavirin is an Inhibitor of Human Immunodeficiency Virus Reverse Transcriptase" Molecular Pharmacology, **1990**, *38* (2), 766-770; and the '315 patent.

The basis for the Examiner's rejection is that mycophenolic acid, ribavirin, and DXG/DAPD are known compounds, as set forth by ICHIMURA, FERNANDEZ-LARSSON, and the '315 patent, respectively, for the treatment of HIV; therefore, it is considered *prima facie* obvious to combine such compounds.

However, to establish a *prima facie* case, the PTO must satisfy three requirements. First, the prior art relied upon, coupled with the knowledge generally available in the art at the time of the invention, must contain some suggestion or incentive that would have motivated the skilled artisan to modify a reference or to combine the references. See In re Fine, 837 F.2d 1071, 1074, 5 U.S.P.Q.2d 1596, 1598 (Fed. Cir. 1988); In re Skinner, 2 U.S.O.Q.2d 1788, 1790 (Bd. Pat. App. & Int. 1986). Second, the proposed modification of the prior art must have had a reasonable expectation of success, determined from the vantage point of the skilled artisan at the time the invention was made. See Amgen, Inc. v. Chugai Pharm. Co., 927 F.2d 1200, 1209, 18 U.S.P.Q.2d 1016, 1023 (Fed. Cir. 1991). Lastly, the prior art reference or combination of references must teach or suggest all the limitations of the claims. See In re Wilson, 424 F.2d 1382, 1385, 165 U.S.P.Q. 494, 496 (C.C.P.A. 1970).

Applicants do not disagree with the Examiner that it is well known to those of ordinary skill in the art that combination therapies for HIV infection are widely available. However, relating back to the parameters of the requirements for a *prima facie* case of obviousness, the

Examiner must not only show suggestion or motivation to combine the references but must show reasonable expectation of success, determined from the vantage point of the skilled artisan at the time the invention was made and the prior art reference or combination of references must teach or suggest all the limitations of the claims. While it is known that drug-resistant variants of HIV can emerge after prolonged treatment with an antiviral agent, hence combination therapies can be helpful in the treatment of HIV, it is not known which drug combinations will be efficacious and/or synergistic rather than antagonistic or simply additive. For example, β-L-D4FC rapidly induces a mutation at codon 184 (methionine to valine), resulting in a high level of resistance to 3TC and FTC. In contrast, β-D-D4FC has a different mutation pattern than β-L-D4FC and does not show any significant cross-resistance to 3TC or FTC.

It was unexpectedly found that the combination of an IMPDH inhibitor with a β-D-1,3-dioxolanyl purine have beneficial effects against HIV beyond simple additive effects. It also was unexpectedly found that the combination of an IMPDH inhibitor with a β-D-1,3-dioxolanyl purine have beneficial effects against drug-resistant HIV, such as DAPD and/or DXG resistant HIV.

Unexpected Results for the Combination of an IMPDH inhibitor with a β-D-1,3-Dioxolanyl Purine for the Treatment of HIV

In the specification, Applicants disclosed biological data regarding the unexpected discovery that the combination of an IMPDH inhibitor with a β-D-1,3-dioxolanyl purine exhibit synergistic effects.

For example, on pages 30-31, combination assays were performed using varying concentrations of DAPD, DXG, Abacavir and AZT, alone or with a fixed concentration of

ribavirin (RBV), in the MT2 cell line. EC_{50} values for the compounds were determined in the presence and absence of 1, 5, 10 and 20 μM RBV (see **Table 2**).

Table 2. EC_{50} values (μM) of RBV with DAPD, DXG, Abacavir, or AZT in MT2 cells

Compound	Control	1 μM RBV	5 μM RBV	10 μM RBV	20 μM RBV
DAPD	18.5 (8)[a]	8.2 (2)	2.9 (2)	1.6 (4)	1.3 (4)
DXG	2.65 (8)	2.05 (2)	0.58 (2)	0.5 (2)	0.22 (2)
Abacavir	4.7 (6)	ND	6.9 (2)	6.4 (4)	5.7 (4)
AZT	1.7 (6)	2.9 (2)	4.6 (2)	5.9 (4)	>10 (4)

[a] = number of replicates

In the MT2 cell line, RBV was not active against HIV replication. Addition of 1, 5, 10 and 20 μM RBV decreased the EC_{50} values obtained for DAPD and DXG. **Table 3** on page 31 of the specification, illustrated the fold differences in EC_{50} values obtained for each of the compounds in combination RBV.

Table 3. Fold differences in EC_{50} values in combination with RBV in MT2 cells

Compound	1 μM RBV	5 μM RBV	10 μM RBV	20 μM RBV
DAPD	2.25	6.4	11.56	14.2
DXG	1.29	4.57	5.3	12
Abacavir	ND	0.68	0.73	0.82
AZT	0.59	0.37	0.29	<0.17

Addition of 20 μM RBV had the greatest effect on the antiviral activity of DAPD and DXG with a 14.2 and 12 fold **decrease** in the apparent EC_{50} values respectively. Importantly, the addition of RBV had **no effect** (less than 2 fold difference in the apparent EC_{50}) on the activity of Abacavir. Further, addition of 20 μM RBV resulted in a greater than 6-fold **increase**

in the apparent EC_{50} of AZT indicating that the combination is antagonistic with respect to inhibition of HIV. Similar results were obtained with the addition of 1, 5 and 10, μM RBV.

Combination assays were also performed in PBMCs using varying concentrations of DAPD, DXG, Abacavir and AZT alone or with a fixed concentration of RBV, as described on pages 32-33 of the specification. The EC_{50} values determined for the compounds in the presence and absence of 1, 5, 10 and 20 μM RBV (see **Table 5**).

Table 5. EC_{50} values (μM) of RBV with DAPD, DXG, Abacavir, or AZT in PMBCs

Compound	Control	1 μM RBV	5 μM RBV	10 μM RBV	20 μM RBV
DAPD	4.5 (19)[a]	2.26 (4)	0.7 (5)	0.16 (5)	<0.03 (3)
DXG	0.15 (9)	0.075 (3)	0.027 (4)	<0.01 (3)	<0.01 (4)
Abacavir	0.54 (9)	0.2 (4)	0.11 (4)	0.03 (5)	<0.03 (5)
AZT	0.003 (7)	0.0035 (3)	0.0026 (3)	0.0022 (3)	0.0021 (3)

[a] = number of replicates

RBV inhibited the replication of HIV-1 in PBMCs with an EC_{50} of 20.5 μM. Addition of 1, 5, 10 and 20 μM RBV decreased the EC_{50} values obtained for DAPD and DXG. **Table 6** on page 33 of the specification, illustrated the fold differences in EC_{50} values obtained for each of the compounds in combination RBV.

Table 6. Fold differences in EC_{50} values with RBV

Compound	1 μM RBV	5 μM RBV	10 μM RBV	20 μM RBV
DAPD	2	6.4	28	>150
DXG	2	5.6	>15	>15
Abacavir	2.7	4.9	18	>18
AZT	0.86	1.2	1.4	1.4

Addition of 20 μM RBV to DAPD, DXG and Abacavir completely inhibited HIV replication in PBMCs at all the concentrations tested but had little effect on the activity of AZT. Addition of lower concentrations of RBV also had a significant effect on the activity of DAPD, DXG and Abacavir.

Combination assays were performed using varying concentrations of DAPD, DXG, Abacavir, AZT and FTC, alone or with a fixed concentration of mycophenolic acid (MPA) in MT2 cells. See pages 37-38 of the specification. EC_{50} values for the compounds were determined in the presence and absence of 0.25, 0.1, and 0.01 μM MPA (see **Table 8**).

Table 8. EC_{50} values (μM) of MPA with DAPD, DXG, Abacavir, AZT, or FTC in MT2 cells

Compound	Control	0.01 μM MPA	0.1 μM MPA	0.25 μM MPA
DAPD	20 (5)[a]	22 (1)	4.9 (1)	1.2 (5)
DXG	2.1 (5)	2.5 (1)	0.6 (1)	0.2 (5)
Abacavir	2.4 (3)	2.4 (1)	2.4 (1)	1.4 (3)
AZT	0.42 (2)	0.3 (1)	0.8 (1)	0.95 (2)
FTC	0.6 (2)	0.62 (1)	0.62 (1)	0.4 (2)

[a] = number of replicates

In the MT2 cell line, MPA was not active against HIV replication. **Table 9** on page 37 and 38 of the specification, illustrated the fold differences in EC_{50} values obtained for each of the compounds in combination with 0.1 and 0.25 μM MPA.

Table 9. Fold Differences in EC_{50} Values in Combination with MPA in MT2 cells

Compound	0.1 μM MPA	0.25 μM MPA
DAPD	4.1	16.7
DXG	3.5	10.5
Abacavir	1	1.7
AZT	0.5	0.44
FTC	1	1.5

Addition of 0.25 μM MPA had the greatest effect on the antiviral activity of DAPD and DXG with a 16.7 and 10.5 fold **decrease** in the apparent EC_{50} values respectively. Addition of 0.25 μM MPA had **little effect** on the activity of Abacavir and FTC, less than a 2 fold decrease in the apparent EC_{50}, and resulted in a 2.3 fold **increase** in the apparent EC_{50} of AZT indicating that the combination is antagonistic with respect to inhibition of HIV. Similar results were obtained with the addition of 0.1 μM MPA.

Combination assays were also performed in PBMCs using varying concentrations of DAPD, DXG, Abacavir, AZT and FTC alone or with a fixed concentration of MPA (pages 39-40). The EC_{50} values determined for the compounds in the presence and absence of 0.25, 0.1, and 0.01 μM MPA were shown in **Table 11**.

Table 11. EC_{50} values (μM) of MPA with DAPD, DXG, Abacavir, AZT, or FTC in PMBCs

Compound	Control	0.01 μM MPA	0.1 μM MPA	0.25 μM MPA
DAPD	4.1 (4)[a]	0.9 (3)	0.18 (5)	<0.0002 (2)
DXG	0.14 (4)	0.015 (3)	0.006 (5)	<0.0002 (2)
Abacavir	1.2 (4)	1.1 (2)	0.38 (3)	<0.0005 (2)
AZT	0.0031 (3)	0.0026 (3)	0.0021 (3)	0.0017 (3)
FTC	0.011 (3)	0.008 (3)	0.0093 (3)	0.006 (2)

[a] = number of replicates

Mycophenolic acid inhibited the replication of HIV-1 in PBMCs with an EC_{50} of 0.095 μM. CC_{50} value obtained for MPA in these cells were 4.5 μM resulting in a therapeutic index of 47. **Table 12** on page 40 of the specification, illustrated the fold differences in EC_{50} values obtained for each of the compounds in combination with 0.01, 0.1 and 0.25 μM MPA.

Table 12. Fold Differences in EC_{50} Values with MPA

Compound	0.01 μM MPA	0.1 μM MPA	0.25 μM MPA
DAPD	4.6	22.8	>50
DXG	9.3	23.3	>50
Abacavir	1.1	3.2	>50
AZT	1.2	1.5	1.8
FTC	1.4	1.2	1.8

Addition of 0.01 μM MPA **decreased** the EC_{50} for DAPD and DXG but had **no effect** on the EC_{50} values obtained for Abacavir, AZT and FTC (less than 2 fold change in EC_{50}).

Therefore, a direct relationship between the concentration of MPA or RBV and the level of synergy seen in combination with DAPD and DXG was observed. The combination of 0.25 μM MPA with DAPD or DXG produced the greatest results without any cytotoxicity. By contrast, addition of MPA resulted in a no change or even a decrease in the activity of AZT. Little to no effect was noted on the activity of abacavir at any of the MPA or RBV concentrations tested.

The results obtained from the combination studies using HIV-infected MT2 cells are summarized in below.

Effect of MPA and RBV on the activity of NRTIs in MT2 cells

Compound	EC_{50} alone	EC_{50} w/ 0.25 μM MPA	Fold Δ*	EC_{50} w/ 20 μM RBV	Fold Δ
DAPD	20 ± 5.4 μM	1.2 ± 0.6 μM	16.7	1.4 ± 1 μM	14.3
DXG	2.1 ± 0.7 μM	0.2 ± 0.1 μM	10.5	0.18 ± 0.15 μM	11.7
abacavir	2.4 ± 0.4 μM	1.4 ± 0.1 μM	1.7	2.9 ± 1.5 μM	0.8
AZT	0.4 ± 0.2 μM	0.95 ± 0.2 μM	0.4	2.5 ± 0.9 μM	0.17

* EC_{50} alone/EC_{50} in combination

Unexpected Results for the Combination of an IMPDH inhibitor with a β-D-1,3-Dioxolanyl Purine for the Treatment of Drug-Resistant HIV

In the specification, Applicants disclosed biological data regarding the unexpected discovery that the combination of an IMPDH inhibitor with a β-D-1,3-dioxolanyl purine exhibit synergistic effects in drug-resistant variants of HIV.

For example, on page 32, the effect of RBV on the activity of DAPD and DXG against mutant strains of HIV was analyzed (see **Table 4**). The restraint strains analyzed included viruses created by site directed mutagenesis, K65R and L74V, as well as a recombinant virus containing mutations at positions 98S, 116Y, 151M and 215Y. The wild type backbone in which these mutants were created, xxLAI, was also analyzed for comparison. These mutations have been shown to develop *in vitro* when virus was passed in the presence of increasing concentrations of DXG. RBV was tested in combination with DAPD and DXG at a fixed concentration of 20 μM. The mutant viruses tested all demonstrated increased EC_{50} values (greater than four fold) for both DAPD and DXG indicating resistance to these compounds. Addition of 20 μM RBV decreased the EC_{50} values of DAPD and DXG against these viruses. The EC_{50} values determined for DAPD and DXG in the presence of 20 μM RBV were at least 2.5-fold lower than those obtained for the wild type virus. These results were summarized in **Table 4.**

Table 4. EC_{50} values (μM) of RBV with DAPD and DXG: Resistant Virus

Virus Isolate	DAPD	DAPD+RBV[a]	DXG	DXG+RBV
K65R	43.7 (5.5)[b]	0.9 (0.1)	3.9 (5)	0.29 (0.4)
L74V	34 (4)	0.5 (0.06)	4.5 (5.6)	0.25 (0.35)
A98S,F116Y,Q151M,T215Y	>100 (>12)	2.6 (0.3)	16 (20)	0.3 (0.4)

[a] [RBV] = 20 μM
[b] indicates fold difference from WT

When tested against mutant strains of HIV-1, the combination of 20 μM RBV with DAPD or DXG decreased the EC_{50} values of these compounds to less than those observed with wild type virus, i.e. the previously resistant virus strains are now sensitive to inhibition by DAPD and DXG.

The effect of MPA on the activity of DAPD and DXG against mutant strains of HIV was also analyzed (see pages 38-39, and **Table 10**). Again, the restraint strains analyzed included viruses created by site directed mutagenesis, K65R and L74V, as well as a recombinant virus containing mutations at positions 98S, 116Y, 151M and 215Y. The wild type backbone in which these mutants were created, xxLAI, was also analyzed for comparison. These mutations have been shown to develop *in vitro* when virus was passed in the presence of increasing concentrations of DXG. MPA was tested in combination with DAPD and DXG at a fixed concentration of 0.25 μM. DAPD and DXG were active against all of the wild type strains of HIV tested. The mutant viruses tested all demonstrated increased EC_{50} values for both DAPD and DXG indicating resistance to these compounds. Addition of 0.25 μM MPA decreased the EC_{50} values of DAPD and DXG against these viruses. These values determined for DAPD and DXG in the presence of 0.25 μM MPA were similar to those obtained for the wild type virus.

Table 10. EC_{50} values (μM) of MPA with DAPD and DXG: Resistant Virus

Virus Isolate	DAPD	DAPD+MPA[a]	DXG	DXG+MPA
K65R	41 (6)[b]	7.9 (1.1)	4 (5.6)	1.2 (1.3)

Virus Isolate	DAPD	DAPD+MPA[a]	DXG	DXG+MPA
L74V	39 (4.9)	6.5 (0.8)	3.8 (4.2)	1 (1.1)
A98S,F116Y,Q151M,T215Y	85 (6)	7 (0.5)	16 (8.4)	1.4 (0.7)

[a] [MPA] = 0.25 μM
[b] indicates fold difference from WT

When tested against mutant strains of HIV-1, the combination of 0.25 μM MPA with DAPD or DXG decreased the EC_{50} values of these compounds to less than those observed with wild type virus, i.e. the previously resistant virus strains are now sensitive to inhibition by DAPD and DXG.

Therefore, both RBV and mycophenolic acid, when combined with DAPD or DXG gave an unexpectedly strong synergistic anti-HIV response against wild-type virus. When tested against wild-type virus, using either human PBMC or MT2 cells, both MPA and RBV decreased the EC_{50} value for DXG by at least 10-fold. In contrast, both MPA and RBV increase the EC_{50} value for AZT and had little or no effect on the activity of abacavir or FTC when tested in the MT2 cell line.

When cells infected with mutant virus resistant to DAPD or DXG were exposed to these combinations, the EC_{50} for DAPD and DXG reverted to wild-type values. MPA and RBV completely reversed the resistance to DXG observed with HIV isolates containing the L74V, K65R, or Q151M mutations, which confer partial resistance to DAPD and DXG. When tested against LAI, addition of MPA or RBV resulted in a greater than 10-fold increase in the anti-HIV activity of DXG. MPA, at a concentration of 0.25 μM, completely inhibited HIV replication in PBMC when combined with DXG. This concentration is below that required to inhibit T-cell proliferation. Similarly, when tested against a mutant virus fully resistant to inhibition by DXG (K65R/Q151M, EC_{50} = 80 μM) MPA and RBV reduced the EC_{50} for DXG to within 5-fold of wild type.